Figure S1. Confirmation of *TAP2*-KO in TISI cell line subclones. *TAP2* gene-knockout-cells, performed by CRISPR/Cas9 method, were isolated HLA-class-I intermediate population and cloned using cell sorter (FACSAria, BD Biosciences). In each clone, cleavage sites and frameshift of the gene were determined by Sanger DNA sequencing by primers in Table SII. (A) PCR condition and agarose gel electrophoresis of *TAP2* gene KO site in the individual clone. Lane number indicates the isolated clone number. PCR fragment of *TAP2* wild site was 390 bp. (B) *TAP2* wild-type gene sequence and the KO alleles' cleavage sites of clone #13 were aligned for comparison. *TAP2* alleles cleavage sites were 17 nucleotides deletion and 22 nucleotides deletion each. Underlines indicate cleavage sites. N, nucleotide not identified; *RC, reverse complement.

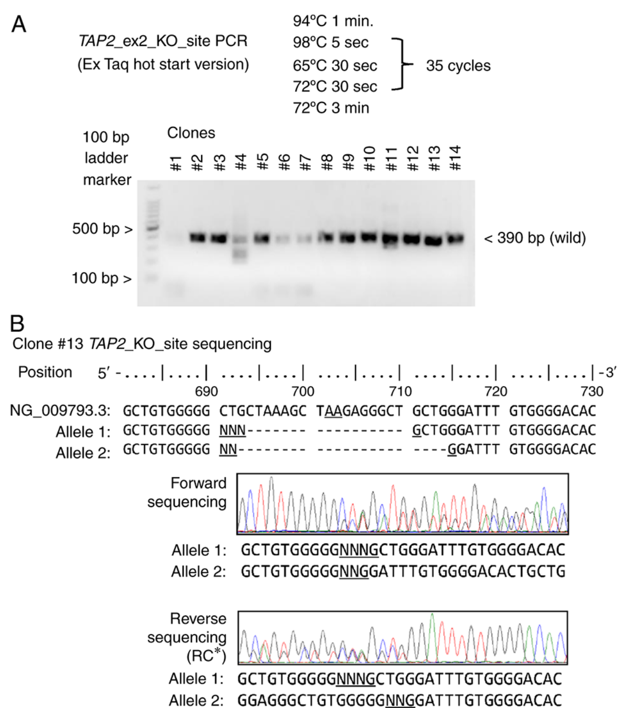

Figure S2. Confirmation of *HLA-A* and *-B* KO in *TAP2*-KO clone. *HLA-A* and *-B* knockout cells, derived from *TAP2*-KO clone #13 shown in Fig. S1, were isolated HLA-class-I low population and cloned by cell sorter (FACSAria, BD Biosciences). In each clone, cleavage sites and frameshift of the gene were determined by Sanger DNA sequencing by primers in Table SII. (A) PCR condition and agarose gel electrophoresis of *HLA-A* and *-B* KO sites in the individual clone. PCR fragment size of the *HLA-A* or *-B* wild site were 589 bp or 556 bp respectively. (B) *HLA-A* gene and (C) *HLA-B* gene cleavage sites DNA sequences of subclone #6-4C. *HLA-A* alleles cleavage sites were 4 nucleotides deletion and 13 nucleotides deletion each (B), and *HLA-B* allesles cleavage sites were 1 nucleotides deletion and 1 nucleotides insertion each (C). Underlines indicate cleavage sites. *RC, reverse complement.

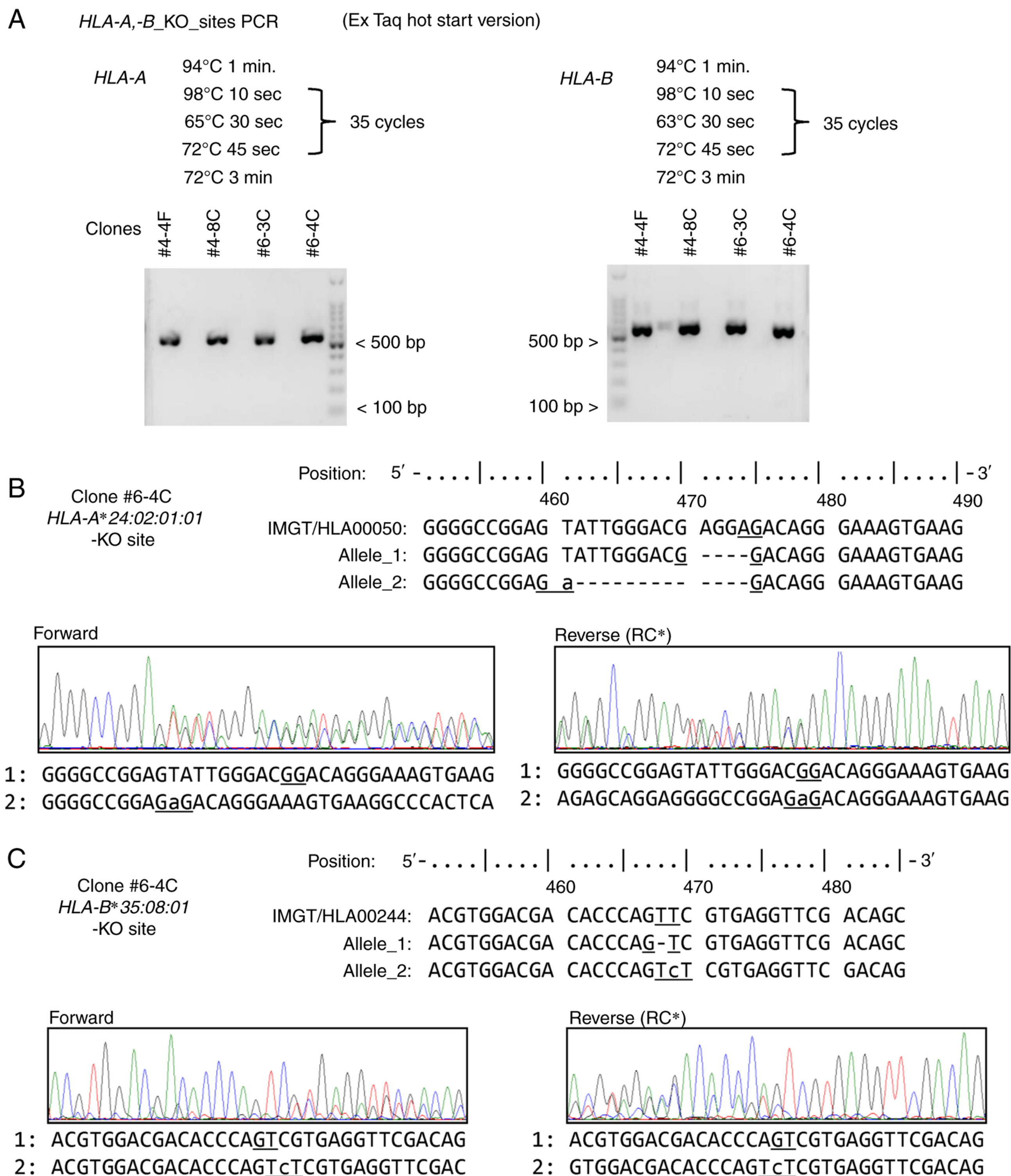

Figure S3. Confirmation of *HLA-C* KO in *HLA-A*, *-B* and *TAP2* KO clone. *HLA-C* knockout cells, derived from clone #6-4C shown in Fig. S2, were isolated HLA-class-I negative population and cloned by cell sorter (FACSAria, BD Biosciences). In each clone, cleavage sites and frameshift of the gene were determined by Sanger DNA sequencing by primers in Table SII. (A) PCR condition and agarose gel electrophoresis of *HLA-C* KO sites in the individual clone. PCR fragments of the *HLA-C* wild site were 591 bp. (B) *HLA-C* gene cleavage sites sequences of subclone #3-9F. *HLA-C* alleles cleavage sites were 2 nucleotides deletion and 7 nucleotides deletion each. Underlines indicate cleavage sites.

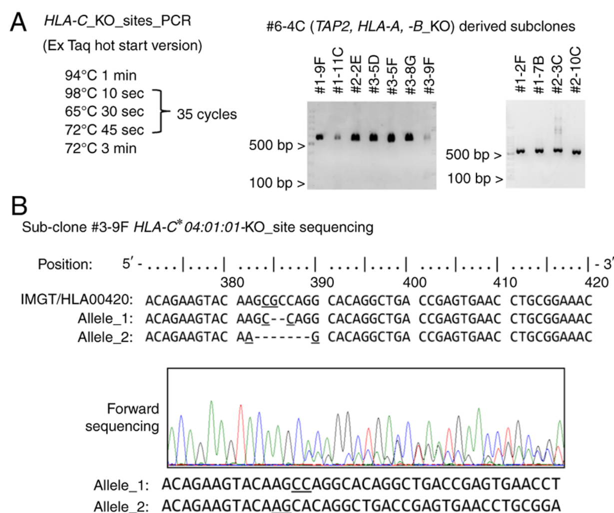

Figure S4. HLA-A allele cDNA knock-in clone screening using PCR and agarose gel electrophoresis. Genomic region between exon 1 and exon 7 of *HLA-A*24:02:01:01* KO loci in the subclone #3-9F shown in Fig. S3 was replaced by other HLA gene allele cDNAs using CRISPR/Cas9 method. The HLA-class-I positive rare cell population were isolated and cloned using cell sorter (FACSAria, BD Biosciences), and the cDNA knock-in clones were determined by HLA-A loci PCR with primers in Table SII and agarose gel electrophoresis. PCR condition for the HLA-A loci and agarose gel electrophoresis image of *HLA-A*33:03:01* or *HLA-A*11:01:01* cDNA knock-in clones as representative were shown. *HLA-A* allele cDNA homo knock-in clone showed 1419 bp single band, and hemi knock-in clone showed two bands (data not shown). The clones #P5F9 (HLA-A*33:03) and #P8B3 (HLA-A*11:01) with no sequencing error were used for further experiments as below.

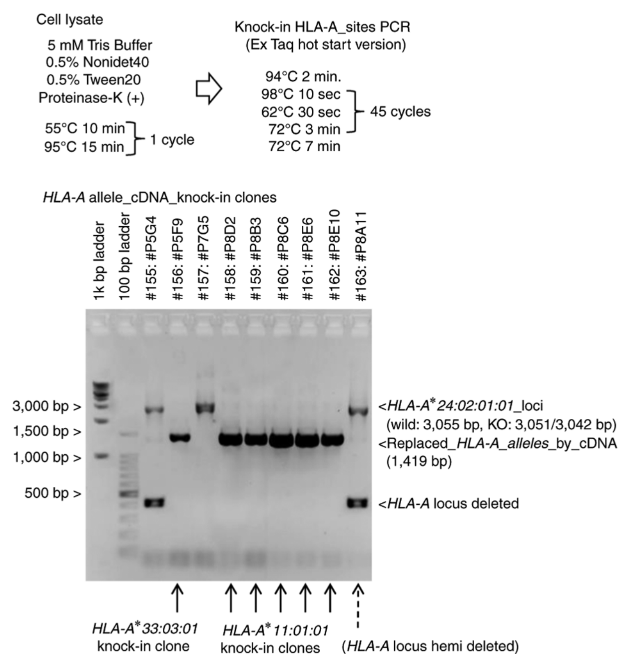

Figure S5. Comparison between the anti-HLA-ABC antibody clone W6/32 and the clone G46-2.6 in T2 cell staining. HLA expression level (x-axis) and cell count (y-axis) were determined by flow cytometry. (A) MHC stabilization assays with immunogenic cytomegalovirus epitope peptides under each culture condition. (B) The assays with synthetic cancer driver mutation-derived epitope peptides. The figure shows representative data from the two experiments. Underlined letters mean AAS positions.